

**Abstracts**  
**Workshop on the Biology of *Kogia***  
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Contents:

**Synopsis of the Anatomy of the Family Kogiidae**

Brian Bloodworth and Christopher D. Marshall

***Kogia* Cardiomyopathy**

Gregory D. Bossart

**Behavior of *Kogia sima* and *Kogia breviceps* Calves in Captivity**

Petra Cunningham-Smith

***Kogia* Distribution in the Northern Gulf of Mexico**

Gregory L. Fulling and Dagmar Fertl

**Recent Progress in the Rehabilitation of Orphaned *Kogia breviceps* and *K. sima* Calves**

Charles A. Manire

**An Approach to the Rehabilitation of *Kogia* spp.**

Charles A. Manire, Howard L. Rhinehart, Nélio B. Barros, Lynne Byrd and Petra Cunningham-Smith

**Examination of Different Methods Used in Age Determination of *Kogia* spp.**

Wayne McFee and Stephanie Plön

**New strandings of pygmy sperm whales *Kogia breviceps* in the NE Atlantic with information on stomach contents**

M. B. Santos, G. J. Pierce, A. López, R.J. Reid & E. Mente

**A Note on the Release and Tracking of a Rehabilitated Pygmy Sperm Whale (*Kogia breviceps*)**

Michael D. Scott, Aleta A. Hohn, Andrew J. Westgate, John R. Nicolas, Brent R. Whitaker and Walton B. Campbell

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## Synopsis of the Anatomy of the Family Kogiidae

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The Family Kogiidae is currently comprised of two species; *Kogia breviceps* (pygmy sperm whale) and *K. sima* (dwarf sperm whale) set apart from Physeteridae based on cranial morphometrics, DNA and soft tissue evidence (Gill, 1871; Schulte, 1917; Handley, 1966; Arnason and Benirschke, 1973; Arnason et al., 1993; Rice, 1998; Ross et al., 2003). Externally, kogiids are counter-shaded dark dorsally and light ventrally with a pronounced crescent shaped “false gill” at the lateral meatus (Schulte and Smith, 1918; Hubbs, 1951; Yamada, 1954; Hale, 1962). The body is robust with a bulbous melon that enlarges with age to produce a “pugged” appearance in older specimens (Hale, 1962; Handley, 1966; Ross, 1978). A discrete, underslung mouth contains 12-16 alveoli in *K. breviceps* and 7-12 in *K. sima*, with the possibility of up to three pairs of erupted maxillary teeth in the later (Handley, 1966; Ross, 1979). Teeth are recurved caudally and fanglike; each tooth is in excess of 32 mm in length (Hale, 1962). Kogiid tooth structure differs from other studied odontocetes in the lack of an enamel cap but are otherwise similar in dentine and cementum layering (Handley, 1966; Ross, 1979). Length at birth is 1.0 m and 1.2 m for *K. sima* and *K. breviceps*, respectively. Sexual maturity is reached by *K. sima* at 2.1-2.2 m and maximum published size is 2.7 m and 280 kg, whereas *K. breviceps* becomes sexually mature at 2.7-2.8 m and is known to reach sizes of 4.25 m and 408 kg. Dorsal fin height of *K. breviceps* is <5% of total body length (TL) and the anterior insertion is >50% of TL from the tip of the snout. Dorsal fin height of *K. sima* is >5% of TL and begins at <50% of TL from the tip of the snout. However, external features are not as accurate as cranial morphometrics, which are more reliable for species identification (Hale, 1963; Handley, 1966; Ross, 1979).

The kogiid skull exhibits the greatest degree of asymmetry of all cetacean skulls (Ness, 1967) and possibly all mammals. This asymmetry is typified by the deviation of the mid-facial sagittal crest, which curves left from the midline (Wall, 1851; Schulte, 1917; Hale, 1963). The nares are also highly asymmetrical; the left side is seven times larger than the right (Schenkkan and Purves, 1973). Numerous additional ossified and soft tissue characters are asymmetrical (Arvy, 1977). The hyoid apparatus contains enlarged, ossified basihyal and thyrohyals (Hale, 1962) to accommodate the mylohyoideus, styloglossus and sternohyoideus musculature (Schulte and Smith, 1918), presumably for suction feeding. The axial skeleton has seven-fused cervical, 12-14 thoracic, 9-12 lumbar and 23-27 caudal vertebrae with 11-18 chevrons (Le Danois, 1911; Yamada, 1954; Hale, 1962, 1963). Vertebral epiphyses remain unfused in *K. breviceps* up to 1.93 m. They begin to fuse at 2.73-2.9 m, and are completely fused by 2.98 m (Hale, 1962). Ribs can be asymmetrical in number, commonly unarticulated caudally, and number from 12-14 (Hale, 1962, 1963). Sternal ribs are cartilaginous, attaching to only three elements of the sternum (Handley, 1966). Inominant bones are absent and replaced by a dense lamella of fibrous tissue providing attachment for penile and abdominal musculature (Schulte and Smith, 1918).

Propulsive epaxial and hypaxial muscle groups are massive and complex, with many large muscles joining and breaking apart (Schulte and Smith, 1918). Coloration is dark red to nearly black, presumably due to a high myoglobin content (Barros, pers. comm.). Intercostals, rectus abdominus and transversalis muscles are well developed (Schulte and Smith, 1918). In the axillary region, the trapezius and all flipper extensors are absent except the common digital extensor while latissimus dorsi, supraspinatus and flexor carpi ulnaris are reduced. Deltoideus and flexor digitorum ulnaris are enlarged and the rhomboideus is well developed with three

heads. Facial musculature are highly undifferentiated and diffuse within fibrous tissue. Narial musculature dilates and retracts the blowhole, and is highly asymmetrical due to the underlying cranial morphology.

A spermaceti organ, located posterior to the melon, is composed of wax esters and triglycerides (Karol et al., 1978). The melon is composed of a denser inner melon of mostly wax esters and an outer melon of mainly triglycerides, although both lipids are common to both tissues. A fibrous coat (junk case) surrounds the spermaceti organ and melon. Sound production is thought to occur as air is moved into a vocal chamber of the right nare and passed through a vocal valve, the “monkey lips” or museau de singe (Clarke, 2003). Presumably, the valve produces sound by the vibration of the vocal reed, a thickened portion on the valve’s dorsal lip edge. Vibrations are transferred to the tip of the spermaceti organ, which focuses sound waves into a beam that passes through the melon and skin. A cushion composed of ligament-like fibers encompasses the vocal valve, and may assist in sound production. A gas recycling system may also be present to prevent air from escaping through the blowhole.

The brain appears similar to other odontocetes, although the separation between dorsal aspects of the orbital lobes is wider than in any other cetacean thus far studied (Marino et al., 2003). Peripherally, Schulte and Smith (1918) identified right and left stellate ganglia innervating each flipper, a condition present in mysticetes. This differs from odontocetes, which possess only one plexus innervating the flipper of *Phocoena* (Schulte and Smith, 1918). This is suggestive of a closer phylogenetic relationship to mysticetes (Schulte and Smith, 1918). The kogiid eye lacks ciliary muscles, has a frail sclera-ciliary body attachment and a relatively low number of retinacular receptor nuclei (Dawson, 1980). However, ample light gathering ability is available from a significant tapetum layer and an optic nerve containing 1.2 million axons, an order of magnitude more than in *Tursiops truncatus*, suggesting good vision in low light environments.

Few investigations of kogiid digestive anatomy are available. The kogiid stomach is comprised of three chambers and one connecting channel (Kernan and Schulte, 1918; Rice and Wolman, 1990). Data from a single, previously frozen sample describes the forestomach as non-glandular and lacking a sphincter to the adjacent fundic chamber. The fundic stomach is glandular and has the potential to reflux digestive fluid into the forestomach. A small connecting channel passes from the fundic to a glandular pyloric chamber, which then opens to a duodenal ampulla and duodenum. An ileum, jejunum and colon are present but a cecum is lacking. However, the composition of biliary bile acids indicates that anaerobic fermentation is occurring in the gut (Hagey et al., 1993). The pancreas resembles that of other mammals, with the exception of A- and B-cells associated with pancreatic acinar cells (Brown et al., 1988). The posterior colon is enlarged into an “ink sac” containing notably liquid brownish-red feces (Benham, 1901; Caldwell and Caldwell, 1989) that can be excreted in large amounts when the animal is startled or excited (Manire et al., 2004), possibly serving as a cryptic screen (Scott and Cordaro, 1987).

Other visceral observations include the heart, lungs and testes. The heart and lungs of kogiids are a smaller percentage of overall body mass than in other cetaceans (Bossart et al., 1985). Testicular sizes for adult males can be large, with lengths up to 0.5 m and masses in excess of 3 kg each testis from a 3 m specimen (Caldwell and Caldwell, 1989).

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# ***Kogia* Cardiomyopathy**

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A renewed interest in the pathologic findings of stranded pygmy (*Kogia breviceps*) and dwarf (*Kogia sima*) sperm whales occurred in 2004 when an increased number of these species stranded along the southeastern United States. Consistent gross lesions reported in the 2004 strandings included emaciation, advanced gastrointestinal nematodiasis, various pulmonary lesions, subcutaneous abscesses, with or without nematodiasis, and cardiomyopathy (CMH).

Briefly, in terrestrial mammals, the identified types of CMH include: “stress”-mediated (an acute process mediated by catecholamines which may lead to sudden death, usually without a history of pre-existing heart disease), hypertrophic, dilated (congestive), restrictive and mixed forms. While each form of CMH is fundamentally different, they are not necessarily mutually exclusive in a given case. Moreover, transitions from one type to another may occur in humans, reflecting chronicity and/or severity of the basic disease process. Bossart et al. (1985) first reported a dilated form of CMH in *Kogia* in 1985. Grossly, a dilated flabby right ventricle, generalized myocardial pallor and chronic passive congestion of the liver were found in the whales of this study. Microscopic lesions of the heart consisted of cardiomyocyte degeneration, loss of cross-striations, interstitial edema and fibrosis. Death was speculated to have been a result of right-sided congestive heart failure. No indication of inflammatory or infectious disease was present. The etiology of the dilated CMH could not be determined but nutritional etiologies were postulated including a thiamine deficiency. Nothing has been published regarding the dilated form of this disease in the peer-reviewed literature since 1985. However, an abstract of a gross and microscopically identical form of CMH in *Kogia* was recently published in Taiwan (Chiu et al., 2003).

To further characterize *Kogia* CMH and help identify a possible etiology, a *Kogia* heart dissection manual was recently developed at Harbor Branch Oceanographic Institution (Hensley et al. 2004). The purpose of this manual is to provide suggested directions for the collection, fixation, and method of dissection of heart specimens of *Kogia* to ensure systematic gross examination and data recording, and the collection of a consistent and specific suite of heart tissue for microscopic study. The manual is intended to meet the needs of the National Marine Fisheries Service Marine Mammal Stranding Network in the ongoing investigation of *Kogia* CMH. Additionally, the manual was designed for use by paramedical/technical personnel who may be less familiar with standard anatomical nomenclature than are professional anatomists. It is for that reason that liberal uses of ordinary terms of reference were employed in the manual. *In situ* examination of the heart is detailed in the manual, which also emphasizes the importance of accurately determining the weights and specific measurements of the hearts. Briefly, for the CMH microscopic study, we recommend four cross sections of the heart. Cross-sections are referred to as levels 1 through 5, from apex to base, respectively. Microscopic evaluation involves the collection of twelve heart sections- septal summit (2 blocks), dorsal wall of right ventricle at level 2, ventral wall of right ventricle at level 2, dorsal wall of left ventricle at level

2, ventral wall of left ventricle at level 2, interventricular septum at level 2, dorsal wall of right ventricle at level 4, ventral wall of right ventricle at level 4, dorsal wall of left ventricle at level 4, ventral wall of left ventricle at level 4, and interventricular septum at level 4 (see Hensley et al., 2004).

Preliminary histopathologic examinations of heart tissue collected according to the new manual protocols were completed in 10 *Kogia breviceps* that had gross and microscopic lesions consistent with CMH. Myocardial lesions considered both acute and chronic were observed in this preliminary study. Lesions seen uniformly in all sections included myofiber cytoplasmic hypereosinophilia and vacuolization, loss of cross-striations, interstitial edema and fibrosis, anisokaryosis with karyomegaly, myofiber disarray (architectural disorganization) and wavy attenuated myofibers. Vacuolization was especially prominent in the sections of heart from the ventricular septum.

Based on this new study, it appears that *Kogia* CMH is best defined as a “mixed form” having components of the stress, dilated and hypertrophic types. Thus, the etiology of CMH in *Kogia* is likely complex and multifactorial. Etiologic components may include metabolic factors such as the excessive repeated sublethal episodes of catecholamine release (repeated acute “stress” reactions) and endogenous glucocorticoid release (chronic “stress” response); transient myocardial ischemia; nutritional deficiencies (e.g., thiamine, alpha tocopherol, selenium); and genetic factors. Additional pathologic studies using the newly designed protocols are needed to confirm these speculations.

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## **Behavior of *Kogia sima* and *Kogia breviceps* Calves in Captivity**

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### **Introduction**

Pygmy sperm whales (*Kogia breviceps*) and dwarf sperm whales (*Kogia sima*) are small, elusive whales that occur in the warm waters along the Atlantic coast of the United States. These smaller relatives of the sperm whale are unusual in appearance, with a shark-like head, false gill markings and robust bodies. Little is known about the most basic aspects of their biology and behavior in the wild, as they tend to avoid boats and are difficult to observe and identify from the air. Most of what is known about this species comes from the investigation of stranded animals. Pygmy sperm whales are the second most common species of cetacean to strand in the southeastern United States after bottlenose dolphins (*Tursiops truncatus*; Odell, 1991). Unfortunately, despite many attempts to rehabilitate these animals, few survive to be released back into the wild. Typically, these animals tend to die within the first few hours or days after rescue. Attempts to maintain these species in aquaria have likewise proved to be unsuccessful (Sylvestre, 1983).

This lack of survival under captive conditions has made behavioral studies of these species difficult. Those adult animals that do survive for short periods tend to be poor subjects for behavioral study, as they are usually mortally ill. Younger animals and calves that strand with their mothers tend to be in better shape physically, will swim and dive more readily, and tend to survive for longer periods (Manire et al., 2004).

At Mote Marine Laboratory's Dolphin and Whale Hospital in Sarasota Florida, we have had the opportunity to observe several *Kogia breviceps* calves and juveniles and one *Kogia sima* calf under captive conditions for periods up to 21 months. Although all of these calves eventually died, we were able to observe some of them for many months longer than either of these species had previously survived in captivity, including weeks and months during which the animals exhibited periods of relatively good health. During the captive period, the calves ranged in age from 1-2 days old to approximately 22 months of age. The *K. breviceps* calves averaged 256 days of care at the hospital, with one individual calf surviving for 631 days. The single *K. sima* calf admitted to the hospital survived for 465 days.

### **Feeding Behavior**

With calves younger than 6 months of age at stranding, it was necessary to use an artificial milk formula to meet all of the animal's nutritional needs. A formula based on a successful recipe used for dolphin calves, modified to meet the nutritional needs of *Kogia* species, has been used at the Dolphin and Whale Hospital to raise orphaned pygmy and dwarf sperm whale calves (Manire et al., 2004). The formula was delivered to the calves via stomach tube. A number of attempts were made to encourage the calves to nurse from a variety of bottles and nipples, but for the most part these were unsuccessful. The anatomy of the *Kogia* spp., particularly the placement of the short, narrow mouth and under slung lower jaw, make it exceptionally difficult for these calves to nurse from a bottle the way other small cetaceans might. To counteract these difficulties, the calves were conditioned to voluntarily approach a handler and receive liquid nourishment by allowing a stomach tube to be passed down their

esophagus. This was first accomplished with handlers in the water; later, some of the calves were conditioned to approach a feeding station at the side of the tank, and the handler could pass the tube into the mouth and through the esophagus while standing outside the tank. Once this behavior was learned, calves appeared to be eager for formula, approaching quickly and assuming a nursing position as soon as the handlers entered the water.

As the calves progressed to 6-7 months of age, they were offered squid both in the water column and by hand from a handler. Calves spent a considerable amount of time mouthing, carrying and playing with the squid before consuming it. One *K breviceps* calf also learned to swallow small fish and fish fillets from a handler.

## Swimming and Diving Behavior

During rehabilitation, all calves were housed in 55,000-gallon rehabilitation tanks in less than 10 feet of water. Such dimensions were clearly not large enough to test the calves' abilities to dive and swim. However, most of the calves were agile swimmers. In spite of reports in the literature that these animals are slow and sluggish (Handley, 1966), the calves at Mote often displayed sudden, startling bursts of speed. Breaching during play was common, and very high breaches (high enough for the vertical animal's tail to clear the water by several feet) were also observed. Calves tended to bask on the surface during resting periods, although slow swimming during periods of obvious rest was also observed. Calves often swam comfortably upside down and engaged in "barrel rolls", with one *K. sima* calf commonly engaging in such behavior even during tube feeding.

When startled or during play, calves of both species would display sudden bursts of speed, emit a cloud of fecal material, then turn around and appear to conceal themselves in the cloud. This behavior occurred even in very young calves, one to two days old. This behavior has also been observed in older animals and may be a mechanism for predator avoidance or capturing prey (Scott et al., 2001).

Before husbandry training, calves tended to avoid capture for medical exams by diving to the bottom of the tank and remaining there until they needed to surface for respiration. From this we were able to record longest breath hold times in both species of calves. The longest breath hold recorded in this manner for a *K. breviceps* calf was 17.33 minutes and the longest breath hold for the *K. sima* calf was 21:40 minutes (Anderson et al., in prep).

## Social Behavior and Husbandry Training

All calves were housed alone during rehabilitation, except two juvenile *K. breviceps* calves that were placed together for about 14 days, and then separated. Little social interaction was observed between these two calves, a male and a female, during the time they were maintained together. Young dependent calves were assumed to be unreleasable, and no effort was made to keep them from becoming habituated to human handlers. *K. breviceps* calves were extremely social with handlers, especially after the tube feeding behaviors were firmly established. The single *K. sima* calf kept for long-term rehabilitation was noticeably more reticent with humans, although this calf eventually became habituated and interactive with handlers as well.

Basic husbandry training was undertaken with the healthiest and longest surviving calves to facilitate weekly medical examinations and weighing. Training was conducted using operant conditioning techniques and positive reinforcement (Ramirez, 1999). Health issues with some of

the calves made training difficult, but several of the calves readily learned one or more of the following husbandry behaviors:

- Stationing for feeding
- Touch and follow a target
- Target discrimination
- Mouth examination
- Girth measurements
- Body desensitization (blowhole, fluke, pectoral fins)
- Swimming into a stretcher for weights and blood draws

All calves were provided with a variety of environmental enrichment devices. All calves were interactive with a number of the devices. Calves displayed curiosity and great interest in new objects and devices in their habitat, frequently pushing objects with their heads or carrying them in their mouths or on their backs. Calves were calm and generally accepting of divers and others in their tanks to clean or facilitate tank maintenance. Younger calves would often assume the calf-echelon position (common to mother-calf pairs in other cetaceans) with humans in the tank, or follow closely and attentively to human swimmers. Calves were extremely tactile and would solicit stroking and body rubs from human caregivers.

Rehabilitation of orphaned *K. breviceps* and *K. sima* calves presents many challenges to both veterinary and animal care staff. Behaviorally, calves of both species appear to be facile learners and are readily adaptable to training and husbandry practices. More research is needed into the behavior of both species in order to provide an optimum captive environment to rehabilitate these animals.

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## ***Kogia* Distribution in the Northern Gulf of Mexico**

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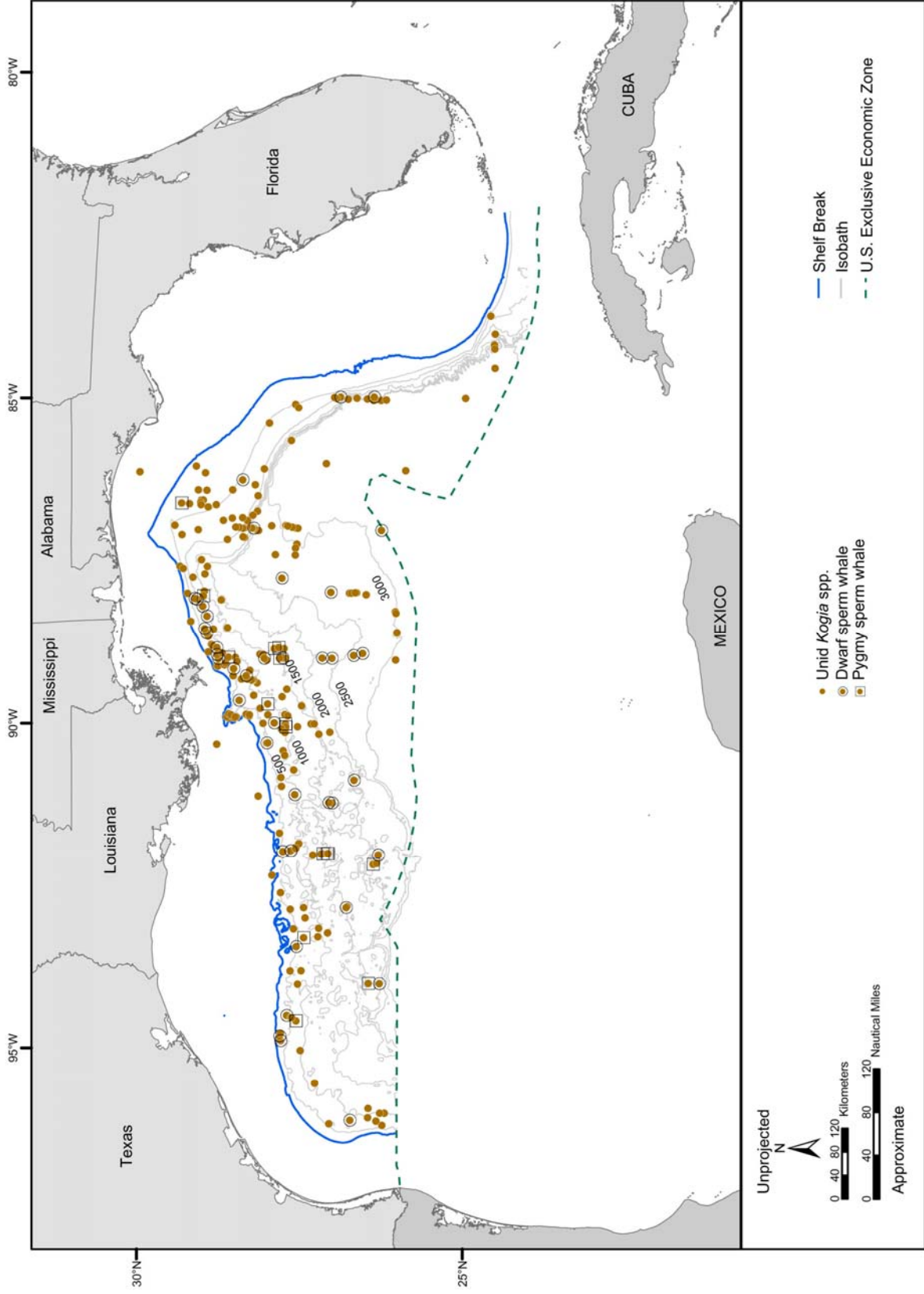
Dagmar Fertl, Geo-Marine, Inc., 550 East 15<sup>th</sup> Street, Plano, Texas 75074 USA

In order to better understand the distribution of *Kogia* in the northern Gulf of Mexico, all available aerial and shipboard sighting records were compiled. Sources included the historical database analyzed in Jefferson and Schiro (1997), as well as shipboard and aerial surveys conducted by the Southeast Fisheries Science Center of the National Marine Fisheries Service (NMFS-SEFSC) and by Texas A&M University.

We compiled a total of 250 sightings (97 aerial and 153 shipboard). Sightings appeared to increase during spring through summer (April-September) reflecting an increase in sighting effort associated with NMFS-SEFSC marine mammal stock assessment surveys, which constitute the majority of the records. It should be noted that these surveys are performed during optimal sighting conditions (e.g., calm seas), and do not represent balanced seasonal coverage throughout any particular year.

It is difficult to identify *Kogia* to species level due to the similarity in appearance and cryptic behavior of both the pygmy and dwarf sperm whale. Seventy-six percent ( $n=191$ ) of all *Kogia* sightings did not have species identity determined (**Figure 1**). *Kogia* predominantly occurred along, and seaward of, the continental shelf break (200 m isobath) (**Figure 1**).

Seventy-six percent ( $n=189$ ) of the sightings were between the shelf break and the 2,000 m isobath; 46% of these ( $n=87$ ) were on the upper continental slope between the 500 and 1,000 m isobaths (**Table 1**). Although there has been little survey effort seaward of the 3,000 m isobath, there are some sightings of individuals in those very deep waters. There is no evidence that *Kogia* regularly occur in continental shelf waters of the Gulf of Mexico (Davis et al. 2000), however, there were some sighting records in waters over the continental shelf (**Figure 1; Table 1**). The shallowest sighting occurred off the Florida Panhandle in waters with a bottom depth of 20 m (**Figure 1**).



**Table 1. *Kogia* distribution in the northern Gulf of Mexico relative to bottom depth.**

<b>Bottom depth (m)</b>	<b>Number of Sightings</b>
0-200	4
200-500	43
500-1000	87
1000-1500	33
1500-2000	26
2000-2500	9
2500-3000	22
3000-3500	26
<b>Total</b>	<b>250</b>

There was a noticeable concentration of sightings in continental slope waters near the Mississippi River delta (**Figure 1**). The Mississippi River delta stands out as an important area for cetaceans in the northern Gulf of Mexico, as evidenced not only from recent survey efforts but also a review of historical records (Mullin et al., 1994; Jefferson and Schiro, 1997; Davis et al., 1998, 2002; Baumgartner et al., 2001). It is well known that oceanographic features are important factors determining cetacean distribution as their prey are attracted to the increased primary productivity associated with some of these features (Biggs et al., 2000; Wormuth et al., 2000; Davis et al., 2002). Baumgartner et al. (2001) suggested that *Kogia* may associate with frontal regions along the shelf break and the upper continental slope, since these are areas with high epipelagic zooplankton biomass, which is likely part of the diet of the common prey species of *Kogia*.

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## Recent progress in the rehabilitation of orphaned *Kogia breviceps* and *K. sima* calves

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Although *Kogia breviceps* is the second most common species of cetacean to strand in the southeastern US and fairly common worldwide and its sister species, *K. sima*, is also regularly found stranded, there have been few animals that have been kept alive for more than a week or two. Very frequently, these stranded animals are cow-calf pairs that are recently post-partum with a severely emaciated cow and a relatively more healthy calf. In an effort to expand the knowledge regarding keeping these two species alive in rehabilitation, the Dolphin and Whale Hospital at Mote Marine Laboratory in Sarasota, Florida has made several attempts to raise kogiid calves. In the process much information has been gained regarding digestive limitations and gastrointestinal disorders that affect these two species, as well as refined medical and husbandry techniques that have allowed for calves to be kept alive for up to 21 months.

Between 1994 and 2003, a total of 10 *K. breviceps* (five adults, one juvenile, and four calves) and three *K. sima* (one each adult, juvenile, and calf) were brought in alive to the Dolphin and Whale Hospital. The adults arrived in fair to very poor body condition and were kept alive from a few hours to a maximum of 40 days. All adults were ultimately found to have electrolyte imbalance, gastrointestinal issues, or cardiomyopathy. In contrast to the arrival condition of the adults, the calves generally arrived in fair to good body condition and were kept alive for 3-21 months.

The approach taken to rehabilitate the calves is soon to be published (Manire et al., 2004). The approach involves the use of a recently developed formula to replace the mother's milk, aggressive treatment of all medical issues as they arise, and very diligent observation for developing medical conditions. Every calf was given a full medical examination including blood tests and cytology exam of gastric, fecal, and blowhole contents at least once weekly. In addition, special attention had to be paid to the animals when they were on medication as they were very sensitive to a number of commonly used drugs (Manire et al., 2002).

The formula used on the later calves was developed as a modification of the SeaWorld dolphin formula (Townsend, 1999) after analysis of several milk samples from lactating *K. breviceps*. Unfortunately, all milk samples analyzed were from sick, dying cows, so the analyses were highly variable. Different ingredients were then tested for digestibility and then blood chemistry values were observed closely during use of the formula (Manire et al., 2004). The calves accepted the formula well and gained weight rapidly on the formula. The only problem encountered with the current formula is possible softening of the teeth.

Aggressive treatment of all medical issues that arose and anticipating other medical issues before they arose was essential to keeping the calves alive for extended periods of time. Failure to address medical issues early and aggressively seemed to lead to worsening of conditions, occasionally beyond the possibility of treatment, as well more secondary issues. However, over-treatment can likewise lead to more secondary issues and a fine balance must be achieved.

Such a fine balance can be achieved only through diligent monitoring for developing conditions. Weekly, thorough exams were a critical part of detecting medical issues before they

reached the point of clinical manifestation. Close behavioral observations likewise aided in detecting problems early.

The issues that caused the most problems, often becoming insurmountable, were those related to the gastrointestinal tract. Anatomically and physiologically, the gastrointestinal tract of kogiids is unique among mammals. First, the contents of the entire intestinal tract appear to be liquid. When there is any form to the contents, this is evidence of constipation. Unfortunately, drugs normally used to control constipation in other mammals seem to have very little effect on the kogiid intestine. In our experience, enemas, stool softeners, saline cathartics, and most other drugs used to treat constipation have little or no effect on kogiids. Keeping fluid intake high, usually through regular stomach tubing, seems to be the only preventative that has much effect. The effects of constipation on these mammals can be devastating, including sequelae such as intestinal volvulus, intestinal rupture, intestinal blockage, and impaction. One of these sequelae have been the ultimate cause of death of each of the kogiid calves that underwent rehabilitation at the Dolphin and Whale Hospital. Research is currently underway to determine other ways of preventing and treating the gastrointestinal problems experienced with these whales.

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## **An Approach to the Rehabilitation of *Kogia* spp.**

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### **Abstract**

Pygmy (*Kogia breviceps*) and dwarf (*K. sima*) sperm whales are rarely seen in the wild, but often seem to live-strand, particularly in cow-calf pairs. The rehabilitation of live-stranded individuals of both species has proven to be exceedingly difficult. The few released animals might not have been completely healthy, an alternative chosen due to their poor survival in captivity. The rehabilitation challenges for *Kogia* are numerous because limited knowledge exists regarding even the basic biology of both species. This report provides information derived from the rehabilitation of 13 live-stranded *K. breviceps* and *K. sima* (including five calves) over the last decade at the Dolphin and Whale Hospital at Mote Marine Laboratory and Aquarium in Sarasota, Florida. One *K. breviceps* calf survived for almost 21 months in captivity and one *K. sima* survived for over 15 months, both apparent worldwide records. From these cases we learned that it is critical to provide supplemental fluids in addition to solid food to maintain continuous activity of the intestinal tract, especially if maintained in chlorine treated water, and that digestibility of squid species typically fed to captive marine mammals was poor. Both species appear to be susceptible to adverse reactions to a number of the drugs commonly used during rehabilitation. In addition, an artificial calf formula was developed to provide adequate nutrition for young calves. Finally, gastric and intestinal stasis appears to lead to death in many of these whales in captivity.

## Examination of Different Methods Used in Age Determination of *Kogia spp.*

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### Introduction

Being able to determine the age of an animal is a fundamental requirement in many areas of research and data on age and growth in relation to reproductive parameters present vital information about the potential survival of a given species. The use of age determination of marine mammals, based on the layered structures in their teeth, has become a standard procedure in stock assessment and management decisions as age-specific estimates for fecundity or mortality are used to project population growth. In addition, changes in parameters such as age at sexual maturity can be interpreted as reflecting changes in the population abundance or resource availability.

Age determination of pygmy (*Kogia breviceps*) and dwarf sperm whales (*K. sima*) has proven to be more problematic than in delphinid species and previous attempts to determine the age of *Kogia spp.* were only marginally successful (Ross, 1979). Without reliable age estimates basic life history parameters of both *Kogia* species, such as age at maturity, ovulation rate and maximum lifespan, remain unknown. In addition, increased stranding rates of *Kogia* in the southeastern United States in recent years has prompted research to investigate the causes of mortality, address nutritional concerns, and environmental factors that may influence distribution. One area of concern has been the identification of cardiomyopathy as a significant cause of mortality (Bossart *et al.*, 1985; Hensley *et al.*, 2004), though the etiology is unknown. Knowledge of age of these animals would greatly improve our understanding of pathological conditions, such as cardiomyopathy, as well as other life history parameters.

Here we describe four methods for age determination used on *Kogia* teeth. Two of these yielded good results and were tested independently on specimens from the southeastern United States and South Africa, Australia and New Zealand, respectively. Although none of the methods has been validated, inherent problems concerning access to these animals may prevent such an approach in the near future. Validation of age determination methods used for delphinid species has been possible through the use of known-age animals living in captivity. However, *Kogia* species typically do not survive more than 1-2 years in captivity, limiting the access of known-age animals for comparative studies.

The use of growth layers in age determination has generally been accepted even when they have not been calibrated for a species based on the facts that 1) growth layers are very common and have been identified in most mammalian species examined, 2) the appearance and structure of growth layers have been similar among different species within the same tissues (e.g.

teeth and bone), and 3) in each species for which data are available, growth layers have been calibrated to real time showing the existence of annually occurring layers.

### **Examination of methods used**

#### Method 1: Formic acid etching – specimens from South Carolina, USA

Teeth from 23 *K. breviceps* from South Carolina, USA were used for age determination. Following the technique of Pierce and Kajimura (1980), each tooth was sectioned along its mid-longitudinal axis using a Buehler Isomet low speed saw with a diamond wafer blade. The half sections were rinsed in running tap water for 30 minutes before being placed in 80 ml of 5% formic acid. It remained in the formic acid bath between one to two hours depending on the amount of occlusion of the pulp cavity. Younger animals with less occlusion required less time in formic acid than did older animals. The half sections were briefly rinsed in tap water and then placed in acetone for two to three minutes. They were then air dried for 10-15 minutes. The tooth was rubbed lightly with a graphite pencil, so that the ridges and grooves of the growth layer groups (GLGs) were distinguishable. Using a binocular microscope at 10.5x magnification, the GLGs were counted and recorded for each animal by two observers on three separate occasions. An average of readings was taken on those animals where disagreement in readings occurred.

#### Method 2: Stained thin-sections- specimens from the southeastern United States

Probably the most widely used method for aging small cetacean species is the use of stained thin-sections following Hohn *et al.* (1989). Generally, this method requires a tooth from the mid to back portion of the lower jaw to be thick sectioned along the mid-longitudinal axis on a low-speed Buehler Isomet saw using a diamond wafer blade. The thick section is then decalcified using a rapid decalcifying agent (RDO), sufficiently rinsed, and then thin-sectioned (25µm) on a freezing sledge microtome. The thin sections are then rinsed and placed in a hematoxylin stain and mounted with glycerol on a slide. GLGs can then be read to obtain an estimated age.

This method works well for small delphinids, but is troublesome for *Kogia* because of the high number of accessory layers that are visible within the GLGs, especially in presumed older individuals. Teeth from 17 of the 23 *K. breviceps* used for age determination in Method 1 were used for comparison of the two methods. Generally, this method resulted in a lower estimation of age and the oldest individual was estimated at 19, compared to 33 for Method 1. Ages were comparable up to 5-6 GLGs between the two methods.

#### Method 3: Laser-ablation Inductively Coupled Plasma Mass Spectrometry

This preliminary study was conducted at the Hollings Marine Laboratory, Charleston, South Carolina, to determine the feasibility of using laser ablation inductively coupled plasma mass spectrometry (LA-ICPMS) to age *Kogia* teeth. In LA-ICPMS a small 213 nm wavelength laser spot (10's of microns in diameter) is used to ablate material off a hard, flat sample surface. The resultant cloud of sample material is then swept into the ICPMS with a carrier gas and subjected to multi-element analysis. Application of the technique to aging is a novel idea and involves monitoring differences in trace element patterns or levels which may present as the laser beam is rastered across alternating GLGs. A preliminary study showed that strontium (Sr) levels changed across GLGs. Advantages of the technique are spatial resolution and increased counting accuracy compared to established methods. Also coupling the trace element information with age may provide insight into life history traits. Disadvantages that may hinder the ability to age

teeth are potential trace element heterogeneity across a tooth and the inability to view the entire sample surface. This feasibility analysis is ongoing.

Method 4: Ground and cleared thin-sections- specimens from South Africa, Australia and New Zealand

Teeth from 60 *K. breviceps* and 44 *K. sima* from South Africa were used for age determination. In addition, teeth were also available for 20 *K. breviceps* and one *K. sima* from Australia and 27 *K. breviceps* from New Zealand.

Standard methods used for age determination in delphinids, such as decalcification and etching with formic acid, proved to present unsatisfactory results in *Kogia* (see above). Oosthuizen and Bester (1997) got similarly poor results using thin sections of decalcified teeth stained with haematoxylin for age determination of Cape fur seals *Arctocephalus pusillus pusillus*. Instead they used the following method which was here adapted for *Kogia* teeth.

Teeth for sectioning were selected on the basis of greatest size, least curving (in no more than one plane) and least wear. Due to the curvature of *Kogia* teeth better results were obtained when teeth were ground down from either side to the midline by hand (rather than embedded in resin and sectioned). The teeth were initially ground down to yield a final 300µm thick section along the midline. A Buehler “Isomet” low speed saw was used for grinding. The saw blade was replaced with an aluminium disc onto which disposable discs of waterproof silicon carbide paper were clamped. The catch container of the saw was filled with water through which the water paper rotated at controlled speed. The initial grind down to a thickness of 1300µm was done using coarse water paper with a grit size of P120. Then finer water paper with a grit size of P320 was used to grind the sections down to 500µm. The final grind using water paper with a grit size of P400 was done without the saw, just by hand, down to a thickness of 300µm. Ground sections were stored in absolute alcohol overnight, cleared in xylene for five minutes and mounted on slides using DPX mounting medium. A binocular microscope with transmitted, polarized light was used to examine the sections. The use of the polarizing light microscope assists in overcoming some of the difficulties in distinguishing any regular layering by expressing structural and histological differences as highly contrasted patterns of light (i.e. different colors or shades) (Myrick, 1980).

**Results and Discussion**

Table 1a, b: Age estimates obtained with the methods described above for different populations of *K. breviceps* and *K. sima*, using Method 1 (South Carolina, USA) and Method 4 (S. Africa, Australia, New Zealand)

a) *K. breviceps*

	South Carolina	South Africa	Australia	New Zealand
Maximum age (females)	14	22.4	10	12.5
Maximum age (males)	33	13	16	16+

b) *K. sima*

	South Africa
Maximum age (females)	21.5
Maximum age (males)	17

The pattern of dentine deposition in the two *Kogia* species differs to that observed in most other odontocetes. It has been described as a 'herringbone' pattern by Klevezal' and Kleinenberg (Klevezal' and Kleinenberg, 1967), while it is usually referred to as a chevron pattern by other authors (Ross, 1979; Perrin and Myrick, 1980). It is characterized by each layer of dentine being deposited at an angle to the longitudinal axis of the tooth. The neonatal line in *Kogia* teeth is clearly distinguishable from the rather "fuzzy" appearance of the prenatal dentine. In comparison, the postnatal dentine is more layered in structure. In general, GLGs were composed of a broad opaque band and a narrow translucent band. *Kogia* teeth show quite conspicuous accessory layers as opposed to the teeth of dolphins, which have finer accessory layers. The general description of the histology of longitudinal sections of *K. breviceps* teeth is in agreement with Ross' (1979) results. The prenatal dentine is easily distinguishable from the postnatal dentine by a clear, narrow neonatal line. In addition, the growth layers in the dentine can be distinguished into GLGs. However, it appears that the technique employed in Method 4 may give a better resolution than the one employed by Ross (1979). His sections were of the same thickness or thinner (200µm) than the ones in Method 4 (300µm). Grinding down the teeth to very thin sections may have resulted in loss of resolution and hence inability to distinguish between laminae as he mentioned in his work. In addition, the use of polarized light in Method 4 appears to improve the resolution of the laminae significantly. A similar result is presented for Cape fur seal *A. pusillus pusillus* teeth (Oosthuizen, 1997). This technique uses the refractive properties of the apatite crystals in the teeth of these animals and thus enables distinction between layers, which would not be possible with normal light microscopy. However, the reason why results with stains such as haematoxylin (Method 2) or toluidine blue (as employed by Ross (1979)) did not yield good results for teeth of either *Kogia* species is unclear.

No significant difference was found between the age estimates from Method 4 and the ones made by Ross (1979) on some of the same specimens, which supports the fact that the interpretation of the GLGs was the same by both observers. Further, growth curves established from Ross' (1979) results and those from Method 4 were very similar. The greatest source of error in age determination is the misinterpretation of GLGs and errors are predominantly due to accessory layers and poorly prepared sections (Hohn et al., 1989). Any guide or "model" for estimating age from odontocete teeth must take into account that accessory layers are often a part of the GLG pattern (Hohn et al., 1989) and this is particularly the case with *Kogia* teeth as the number of accessory layers present is high in the two species. This highlights the need to standardize age estimation techniques for *Kogia* in order to compare results from different studies.

It should be kept in mind that for both *Kogia* species the present results on age determination can only be viewed as an estimate. Estimates of age are imprecise for most cetacean species as the definite rate of dentine and cementum deposition is not known for the majority of species. Furthermore, the age estimation is subjective and based on the observers' interpretation of GLGs. This imprecision tends to increase with the age of the animals. Although the present study presents the first age estimates for a comparatively large sample of *Kogia* and thus a good start into exploring the life history strategy of both species, the technique may be refined and perfected once teeth and age estimates from more animals become available. In particular, reading teeth from animals originating from different geographical locations may aid in defining the GLG patterns in the teeth of the two *Kogia* species.

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New strandings of pygmy sperm whales *Kogia breviceps* in the NE Atlantic with information on stomach contents

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Stomach contents were collected from four out of 10 pygmy sperm whales stranded in Galicia (NW Spain) between 1983 and 2003. The first whale, a female 287 cm long, was found at San Xurxo (Ferrol) on 25 October 1995. Three days later, a lactating female stranded at the same location. On 3 October 1997, a pregnant female 324 cm long (with a male foetus of 125 cm) was found at Baldaio. The fourth whale, a 212 cm male was found stranded at Punta Hermida on 27 January 2002. In Scotland, an adult female and her calf stranded in Loch Ryan near Stranraer on 18 October 1999; these whales represent the first record of the species in Scotland. The adult female was 268 cm long and was in the early stages of pregnancy with a male foetus 25cm long. The 208 cm calf originally stranded with its mother but was not recovered until 11 days later. Due to the advanced state of decomposition its sex could not be determined.

Cephalopod beaks in stomach contents were identified using a published guide (Clarke, 1986) and a reference collection of oceanic cephalopod beaks. Standard measurements were taken of the lower beaks. All undamaged lower beaks were measured except in the cases of one of the Ferrol whales and the Baldaio specimen, which contained large numbers of *Histioteuthis reversa* beaks, of which a random sub-sample of 200 was measured in each case. Mantle length (ML) and body weights of cephalopod prey were estimated from lower beak measurements Clarke (1986). The number of individuals of each cephalopod species present in a stomach was estimated from the number of beaks. Fish bones (two mandibles) were identified using reference material. Crustacean remains were in a very poor state of preservation and could not be identified to species level. Overall diet composition for each whale was calculated in terms of prey numbers and biomass by expressing the counts and summed weights, respectively, for each prey species as a proportion of the all-species totals.

Food remains of the Galicean whales (Table 1) consisted almost entirely of cephalopod beaks: 941 upper and 1072 lower beaks in the first whale stranded at Ferrol, 233 upper and 231 lower beaks in the second, 795 upper and 807 lower beaks in the whale stranded at Baldaio, and 12 upper and 22 lower beaks in the whale stranded at Punta Hermida. Fish eye lenses were found in all stomach; two dentaries identified as belonging to Sloane's viperfish *Chauliodus sloani* were found in the stomach of the Punta Hermida whale. Crustacean remains were found in three of the whales. The giant mysid of the genus *Gnatophausia* sp. was identified from the first whale stranded at Ferrol and a small parasitic isopod was identified from the whale stranded at Punta Hermida. The squid *Histioteuthis reversa* was the most numerous prey consumed and comprised around 93%, 82% and 89% of the estimated weight of prey items eaten by the Ferrol whales and the Baldaio whale, respectively. For the whale stranded at Punta Hermida, cephalopods of the family Histioteuthidae comprised more than 40% of the estimated weight of prey, while another cephalopod species, *Lepidoteuthis grimaldi*, made up a further 40%. The estimated mantle lengths of the *H. reversa* eaten ranged from 5 to 65 mm with a modal length of around 45 mm.

Other cephalopod species found in the stomachs included *H. bonnellii*, *Todarodes sagittatus* and *Teuthowenia megalops*.

For the whales stranded in Scotland, again most of the remains consisted of cephalopod beaks: 139 upper and 109 lower beaks in the adult female and 115 upper and 104 lower beaks in the calf. Fish eye lenses were found in both whales, also crustacean remains in the calf stomach. Again squid of the family Histioteuthidae were the main prey. The mature female had eaten mainly *H. bonnellii* (53% of the estimated weight of prey items) and the calf had taken mainly *H. reversa* (43.5% of the estimated prey weight). Estimated sizes for *H. reversa* eaten by the Scottish whales varied between 15 to 45 mm ML with two modes at 25 and 45 mm for the adult female, and from 5 to 45 mm with a mode at 15 mm for the calf.

All the pygmy sperm whale stomachs analysed contained mainly oceanic cephalopod remains and very little else. *Histioteuthis reversa*, the most common prey, is an oceanic squid with a recorded maximum size of 186 mm M and found at up to 1000 m depth (Voss et al., 1998), moving closer to the surface at night and to deeper waters during the day. The other prey remains identified included *Chauliodus sloani*, a mesopelagic and bathypelagic species believed to be quite common in deep oceanic waters (Whitehead et al., 1989). The crustacean remains identified belonged to the genus *Gnatophausia*, a group of giant mysid bathypelagic species. The most common species in the genus, *G. ingens* has been recorded to reach 35 cm in length and to live in waters ranging from 400 to 800 m depth (Barnes, 1974; Sanders & Childress, 1990). The distribution and ecology of the majority of the species in the diet seem to suggest that pygmy sperm whales feed generally in deep shelf and slope waters beyond the continental shelf.

The pygmy sperm whales studied had very similar stomach contents, closely corresponding to the diet of this species recorded elsewhere (e.g. Ross, 1978; Martins et al., 1985; Klages et al., 1989; McAlpine et al., 1997). The main prey (family Histioteuthidae) were also recorded by authors analysing the stomach contents of this species further south in the Atlantic (Martins et al., 1985; Hernández-García, 1995). Cephalopod beaks recovered from three previous European strandings of the species were not identified (Allen, 1941; Duguy, 1966).

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Table 1. Prey species found in stomachs of pygmy sperm whales. For all prey types, number of individuals (N) and importance (%W=percentage by weight) are indicated. For cephalopods, numbers of beaks found are indicated (lower beaks unless otherwise stated). For fish and crustaceans, the type of remains found is indicated. Some *Histioteuthis* beaks could not be identified to species but belonged to the “type A” group (Clarke, 1986), which includes *H. arcturi*, *H. corona*, *H. meleagroteuthis* and *H. bonnellii*.

		Ferrol (287 cm)		Ferrol (266 cm)		Baldaio (324 cm)		P. Hermida (212 cm)		Scotland (268 cm)		Scotland (208 cm)		
Family	Species	N	% W	N	% W	N	% W	N	% W	N	% W	N	% W	
Cephalopod prey	<i>Rossia macrosoma</i>	-	-	-	-	-	-	-	-	-	-	1	2.08	
	Unid. Sepioids	-	-	-	-	-	-	-	-	12	0.43	72	22.39	
Octopoteuthidae	<i>Octopoteuthis sicula</i>	-	-	-	-	2	0.84	-	-	1	0.74	-	-	
Gonatidae	<i>Gonatus</i> sp.	3	0.40	1	0.93	3	0.55	-	-	8	6.58	1	2.37	
Lepidoteuthidae	<b><i>Lepidoteuthis grimaldii</i></b>	-	-	-	-	-	-	1	40.73	-	-	-	-	
Histioteuthidae	<i>Histioteuthis reversa</i>	997	64.50	194	63.91	724	53.09	3	5.17	39	10.68	27	43.49	
	<i>H. bonnellii</i>	16	14.65	6	19.89	7	9.38	-	-	15	53.50	-	-	
	<i>H. meleagroteuthis</i>	-	-	-	-	-	-	1	4.83	-	-	-	-	
	<i>H. type A</i>	13	1.62	1	0.38	37	18.58	6	30.27	3	0.61	3	2.35	
Brachioteuthidae	<i>Brachioteuthis riisei</i>	1	0.01	2	0.14	-	-	-	-	3	0.08	1	0.39	
Ommastrephidae	<b><i>Todarodes sagittatus</i></b>	17	16.81	1	2.60	8	11.14	-	-	2	11.51	1	14.37	
	Unid. Ommastrephids	-	-	1	0.48	-	-	-	-	5	5.35	-	-	
Chiroteuthidae	<i>Chiroteuthis veranyi</i>	1	0.07	-	-	1	0.13	3	4.97	-	-	-	-	
	<i>Chiroteuthis</i> sp. (type 2)	3	0.39	-	-	1	0.26	1	3.36	10	5.23	1	0.33	
Pholidoteuthidae	<b><i>Pholidoteuthis</i></b>	1	0.33	-	-	3	4.22	-	-	-	-	-	-	
Mastigoteuthidae	<i>Mastigoteuthis schmidtii</i>	-	-	1	0.05	1	0.09	3	2.36	-	-	-	-	
Cranchiidae		1	0.04	3	0.59	3	0.16	3	5.43	-	-	-	-	
	<i>Galiteuthis armata</i>	-	-	-	-	1	0.29	1	2.88	-	-	-	-	
	<i>Teuthowenia megalops</i>	14	0.91	21	11.02	16	1.26	-	-	10	2.61	5	12.24	
	<i>Teuthowenia</i> sp. (type 2)	1	0.24	-	-	-	-	-	-	2	2.68	-	-	
Alloposidae	<i>Haliphron atlanticus</i>	1	-	-	-	-	-	-	-	-	-	-	-	
Octopodidae	Unid. Octopod	-	-	-	-	-	-	-	-	-	-	1	-	
Fish and crustacean prey														
Family	Species	N	% W	N	% W	N	% W	N	% W	N	% W	N	% W	
Chauliodontidae	<i>Chauliodus sloani</i>	-	-	-	-	-	-	1	-	-	-	-	-	
Unidentified	Unid. fish	1	-	1	-	1	-	-	-	1	-	1	-	
Mysidacea	<i>Gnatophausia</i> sp.	2	0.04	-	-	-	-	-	-	-	-	-	-	
Isopoda	Parasitic	-	-	-	-	-	-	1	-	-	-	-	-	
Unidentified	Unid. crustacean	-	-	1	-	-	-	-	-	-	-	11	-	
											-	-	-	-
<b>TOTAL</b>		<b>1075</b>	<b>100</b>	<b>235</b>	<b>100</b>	<b>808</b>	<b>100</b>	<b>24</b>	<b>100</b>	<b>152</b>	<b>100</b>	<b>128</b>	<b>100</b>	

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## **A note on the release and tracking of a rehabilitated pygmy sperm whale (*Kogia breviceps*)**

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### ABSTRACT

A stranded, rehabilitated 220cm female pygmy sperm whale was radiotracked from 31 May-4 June 1994 after its release in the Gulf Stream off Cape Canaveral, Florida. The whale moved directly off the continental shelf and headed northward within a corridor bounded by the shelf break and the eastern edge of the Gulf Stream. It moved offshore up to 32 n. miles from the shelf break during the late afternoons and nights and headed back toward the shelf break during the day. The average travelling speed was 3.0kts, and ranged from 0-6kts. Speeds were greatest offshore of the shelf break (4.7kts), where the speed of the Gulf Stream was the greatest, and both travelling speeds and Gulf Stream speeds decreased with distance offshore. The whale did not appear, however, to be drifting passively with the current. Diving duration varied significantly with light levels. The whale made long dives (> 8min) at night and on overcast days when squid are known to be closer to the surface. During clear days, the whale's dives were significantly shorter, typically less than five minutes ( $n = 841$ ). Although these results come from only a single, rehabilitated animal, the four days of data provided the first information on pygmy sperm whale movements and diving behaviour at sea: how its behaviour was influenced by time of day, oceanographic features, and environmental conditions, and how the whale's surfacing behaviour could allow survey estimates to be adjusted for diving whales missed along the trackline.

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